

Supplementary Tables.

Supplementary Table I. Outcome variables calculated for the 15 day period preceding the day of interest.

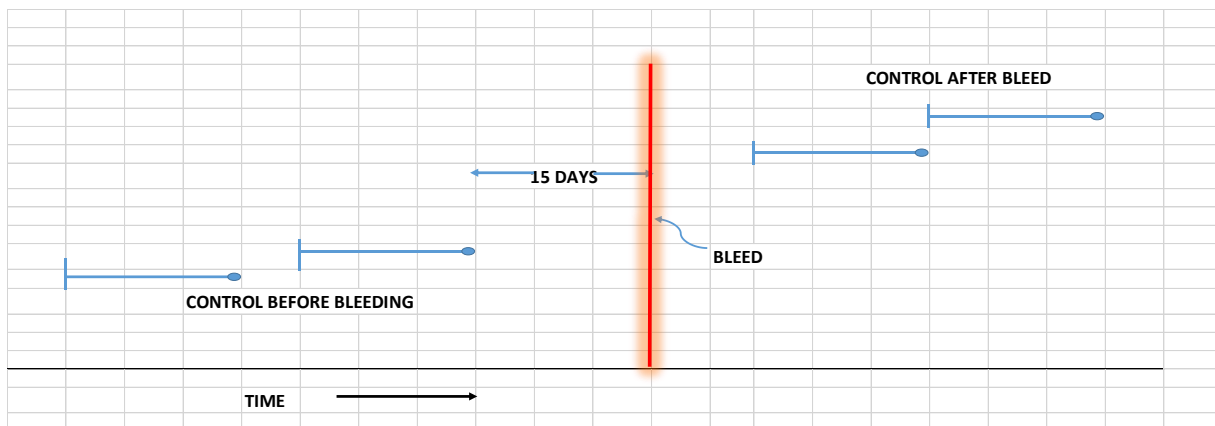
Outcome	Description
Dose	Number of units of factor used for each infusion, taken from each patient's infusion log.
Total amount of factor used	The total number of units of factor infused during the 15-day period, taken from each patient's infusion log.
Area under the curve (AUC)	Calculated from the patient's HL and V_d : $AUC = (Dose * HL) / (V_d * 0.693)^+$
Total number of concentration hours	Factor concentration (units/dL) at the end of each hour, summed over 15 days.
Time below: 5 IU.dL ⁻¹ 15 IU.dL ⁻¹	Hours below each threshold, calculated by the simulation model.

HL, Half-life (hours). V_d , volume of distribution (dL).

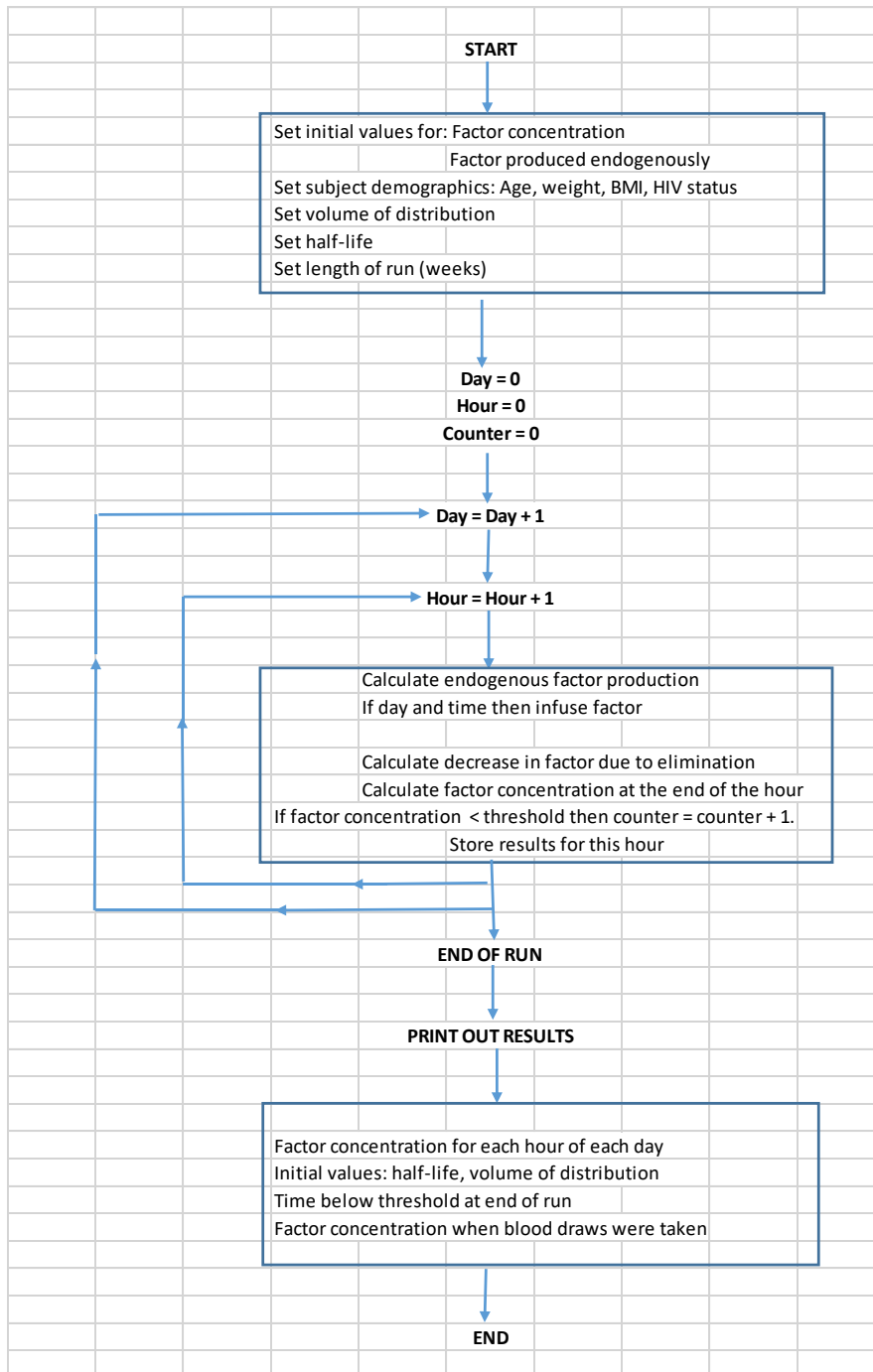
⁺Bauer LA. Applied Clinical Pharmacokinetics. McGraw Hill Medical, New York. 2014;3:868.

Supplementary Figures.

Supplementary Figure 1. Schematic diagram showing the 15 days preceding the bleed, the two control periods before that episode, and the two control periods afterwards.



Supplementary Figure 2. A flow diagram of the pharmacokinetic simulation model that calculates factor concentration for each patient. After setting the initial values for the patient, the model calculates the factor concentration at the end of each hour. The model can be run for any length of time set by the operator.



Simulation model

A simple model simulated the concentration in the blood at each hour of the day. The purpose of this model was to calculate the time spent below threshold levels and the total area under the curve during a period of specified length. Initial values were set: the patients' trough concentration, age, weight, and HIV status. The half-life and volume of distribution was set for that patient as described in the Methods section of the main text. The length of the run was set, while day, hour and all counters were set to zero. The information (day, hour and number of units) for each infusion was read into the program.

The model's 'currency' was the number of factor units in the body, from which the factor concentration was calculated. The model started at 00:01 hours on Day 1. At the beginning of each hour the endogenous factor production was calculated for that hour. Taking the amount of factor in the body after adding endogenous factor production, and applying the half-life, the program then calculated the amount of factor lost by elimination during that hour. It then calculated factor concentration at the end of the hour. The factor level and concentration were stored in memory. The program then stepped to the next hour and ran through the same calculations. After 24 hours it stepped to the next day and cycled through the 24 hours again. Thus there was a cycle of hours embedded in a cycle of days, running for as many days as required (Supplementary Figure 2).

As the program cycled through the hours, if it detected that it was the day and time for an infusion, then the number of units for that particular infusion was added to the level of factor in the blood. At the end of the run the program printed out a summary of the input variables and of the outputs of interest. The comparison between the measured factor concentration and the concentration predicted by the model was a means to validate the model.

This is a single-compartment model. A two-compartment model would be more realistic but requires good estimates of the diffusion coefficients between the two compartments. Lacking such estimates, we have continued with the single-compartment model.

This basic model could be modified with just a few lines of code to calculate any pharmacokinetic parameters of interest. For example, at the end of each hour it determined whether factor concentration fell below, say, 15 IU.dL^{-1} . If yes, then a counter was updated (e.g. $\text{counter} = \text{counter} + 1$). At the end of the run the print-out included a table showing the time spent below 15 IU.dL^{-1} . This could be further modified to print out the number of hours below 15 IU.dL^{-1} during the night (10 pm to 6 am) and during waking hours (6 am to 10 pm). One could select any threshold of interest. We also asked it to calculate the standard AUC as well as the total concentration-hours (unit.hours/dL) which is the summed hourly concentration for 15 days at a time.

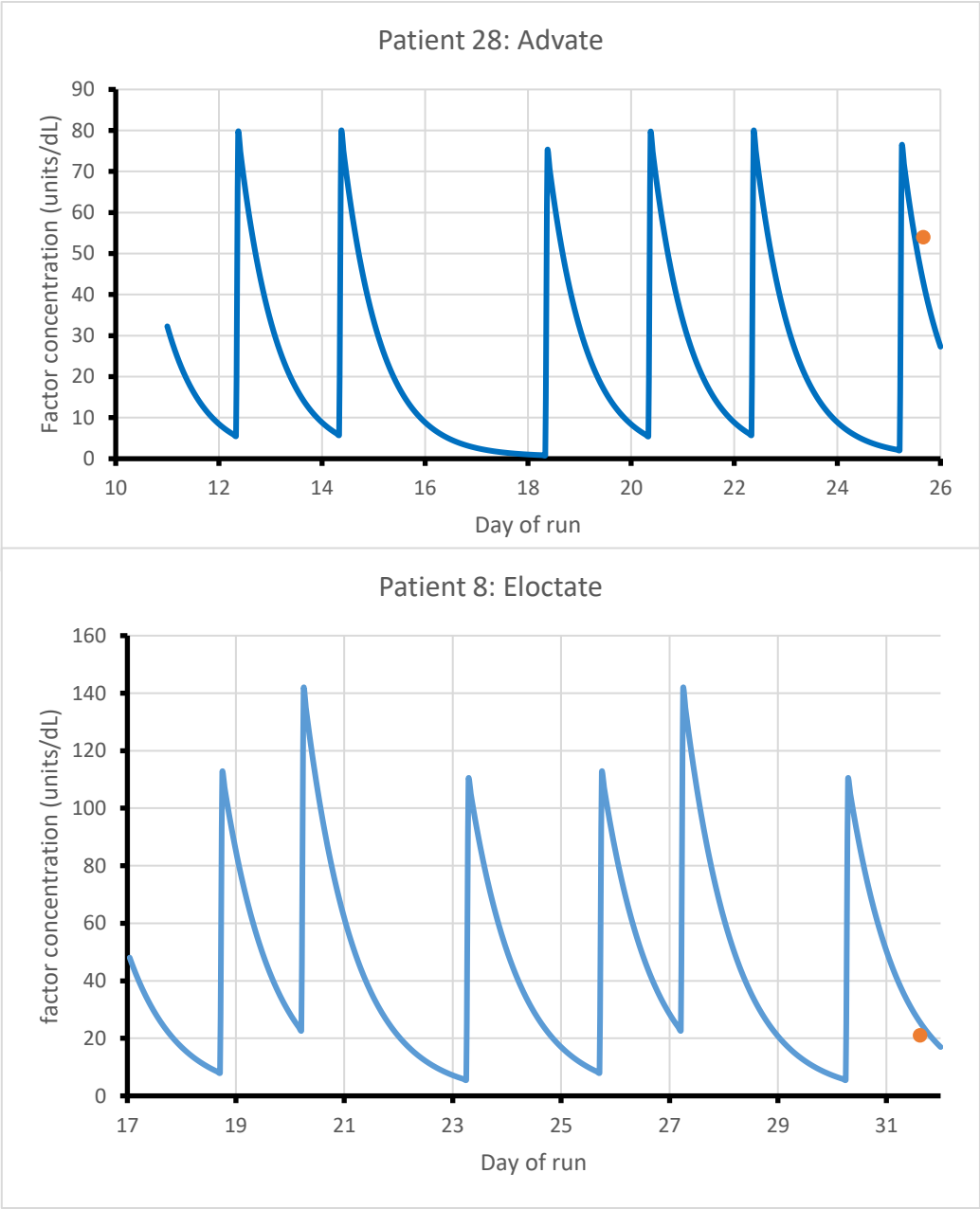
For example, if a patient hypothetically maintained a constant level of 15 IU.dL^{-1} for 15 days, then the total concentration-hours for that period would be $15 * 24 * 15 = 5400 \text{ unit.hrs/dL}$. Since factor concentrations are not constant but vary, concentration-hours reflect exposure to clotting factor better than the AUC as they account for variations in the frequency of infusions. This is important since “real world” infusion logs showed that only some patients infused at regular hours, while many infused at any hour of the day or night. The model also calculated the number of hours spent below the clotting factor thresholds of 5 IU.dL^{-1} and 15 IU.dL^{-1} (see for depiction of the model).

For the 15 days immediately preceding each “day of interest” the model calculated the AUC, the concentration-hours and the time spent below a clotting factor threshold (Supplementary Table I). It

summed the total amount of factor consumed during that period. The model was run for some weeks before the 15 day period to ensure that it had stabilized. Specifically, for episodes from patients with hemophilia A, the model ran for 31 days: a 15 day run-in, the 15 days preceding the day of interest, while the next day (day 31) was the day of interest. For episodes from patients on extended FIX half-life products, the run-in period was prolonged to 60 days.

There were 18 cases where a blood draw coincided with an unbroken run of infusion records. In each case, values from the simulation model and calculated from actual assay levels were compared and found to have very good to excellent correlation in 2/3rds of those tested (12 out of 18). Two representative examples are provided in Supplementary Figure 3.

Supplementary Figure 3. Representative example of values from simulation model and calculated levels from clotting factor assay. The blue line shows values calculated hour by hour by the model (i.e. the predicted values). The red dot is the measured value from the blood draw (i.e. calculated levels from clotting factor assay).



Supplementary Figure 4. Comparison of output from model calculating time spent below factor concentration thresholds of 5 IU.dL⁻¹ and 15 IU.dL⁻¹ for subjects who had their half-lives and volumes of distribution estimated by the WAPPS system (using the NONMEM program) compared to estimates published by the drug company. For those subjects (n=11) who had their half-lives and volumes of distribution estimated by the WAPPS system (using the NONMEM program), we ran the model using those values and noted the output compared to using the half-life and volume of distribution published by the drug company in their inserts. In the graph below, the NONMEM estimates (y-axis) are plotted against the output from the models using the published values of half-life and volume of distribution from the drug company insert (x-axis). The Pearson correlation coefficient (r) is 0.90 and 0.94 for thresholds for 5 and 15 IU.dL⁻¹ respectively.

